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Indigenous microbial capability in solid manure residues to start-up solid-phase anaerobic digesters

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ABSTRACT

Batch solid-phase anaerobic digestion is a technology for sustainable on-farm treatment of solid residues, but is an emerging technology that is yet to be optimised with respect to start-up and inoculation. In the present study, spent bedding from two piggeries (site A and B) were batch digested at total solids (TS) concentration of 5, 10 and 20% at mesophilic (37 °C) and thermophilic (55 °C) temperatures, without adding an external inoculum. The results showed that the indigenous microbial community present in spent bedding was able to recover the full methane potential of the bedding (140 ± 5 and 227 ± 6 L CH_4 $kgVS_{fed}^{-1}$ for site A and B, respectively), but longer treatment times were required than for digestion with an added external inoculum. Nonetheless, at high solid loadings (i.e. TS level > 10%), the digestion performance was affected by chemical inhibition due to ammonia and/or humic acid. Thermophilic temperatures did not influence digestion performance but did increase start-up failure risk. Further, inoculation of residues from the batch digestion to subsequent batch enhanced start-up and achieved full methane potential recovery of the bedding. Inoculation with liquid residue (leachate) was preferred over a solid residue, to preserve treatment capacity for fresh substrate. Overall, the study highlighted that indigenous microbial community in the solid manure residue was capable of recovering full methane potential and that solid-phase digestion was ultimately limited by chemical inhibition rather than lack of suitable microbial community.

1. Introduction

Spent bedding is a lignocellulosic residue that is produced when livestock is housed on straw or similar crop residues (Kruger et al., 2006). Anaerobic digestion (AD) is a suitable treatment option for lignocellulosic residues, with the benefit of renewable energy and the potential for enhanced nutrient recovery (Jha et al., 2011). Spent bedding has unique features for AD, because the manure deposited in the bedding can provide intrinsic bioactivity to pre-ferment the bedding prior to AD (Cui et al., 2011; Tait et al., 2009). For a particulate substrate such as spent bedding, the overall rate of AD is likely dictated by hydrolysis and methanogenesis (Batstone and Jensen, 2011).

Leachbeds (also known as percolation or batch solid-phase digesters) operate at relatively high solids content (>20%) (Batstone and Jensen, 2011), making them particularly suitable for residues such as spent bedding (Yap et al., 2016). Typically, external inoculum would be required for a balanced microbial population during leachbed start-up. However, external inocula may not be available for decentralised on-farm digestion in Australia, because many farms are very remote and have biosecurity restrictions limiting the flow of materials between farms. A small number of studies have examined whether the indigenous microbial community in manure-laden spent bedding could provide self-inoculation (Kusch et al., 2008; Tait et al., 2009). This would be of benefit because the addition of an external inoculum would reduce the available AD capacity for fresh substrate. However, results from a recent study suggested that the lack of indigenous microbial presence could limit the recovery of methane potential from spent bedding in a pilot-scale leachbed (Yap et al., 2016). Previous studies have shown low recovery of methane potential from AD of agricultural wastes in a leachbed (Kusch et al., 2008; Lehtomaki et al., 2008).

Leachbeds may operate at thermophilic conditions for part of their batch life (Pietsch, 2014). That is, when pre-aeration is applied during start-up, temperature is increased by the heat released from pre-composting, thereby reducing the requirements for external heating (Kusch et al., 2008). It has been demonstrated that AD rate can be higher at thermophilic (55–70 °C) than at

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mesophilic conditions (35–37 °C) (De la Rubia et al., 2012; Fernández-Rodríguez et al., 2013). This may be related to increased microbial activity and growth rate at thermophilic temperatures (De la Rubia et al., 2012; Fernández-Rodríguez et al., 2013). Despite enhanced rate kinetics, there have been contradicting reports on the stability of start-up and operation of thermophilic digesters (De la Rubia et al., 2012; Hegde and Pullammanappallil, 2007). Specifically, the start-up of a thermophilic digester can be constrained by a lack of acclimated microbes, leading to process instability and higher risk of failure (De la Rubia et al., 2012). Further, due to the downward shift of the ammonia acid-base pKa with increasing temperature, free ammonia concentration at a given total ammonia concentration increases with increasing temperature. This can also contribute to process instability when treating wastes containing manure (Chen et al., 2008). Nevertheless, there have been limited study of the sensitivity of indigenous microbial community in manure residues towards temperature during digester start-up, even though this may be of general importance for leachbeds.

Another key operational parameter of the leachbed process is the initial solids loading. Loading of rapidly degradable substrate often leads to digester failure (Motte et al., 2013) due to rapid volatile fatty acid (VFA) accumulation and can subsequently lead to inhibition of methanogenic activity due to low pH. Also, high solids loading may elevate inhibitor and/or toxicant concentrations within the leachbed system, thus affecting digestion efficiency (Motte et al., 2013). For instance, when treating nitrogen-rich substrates, such as manure, the increased total ammonia-nitrogen (TAN) concentration that coincides with higher solids loading may lead to free-ammonia inhibition (Wilson et al., 2013; Yenigun and Demirel, 2013). Further, a study by Fernandes et al. (2014) suggested that humic and fulvic acid inhibited hydrolysis. It remains unclear whether the indigenous microbial community in solid livestock residues could appropriately inoculate start-up of a leachbed, despite the high solids loading and the presence of inhibitors.

The aim of the present study was to evaluate the sufficiency of indigenous microbial activity present in fresh spent bedding for solid phase AD. The study examined the response to operating temperature (mesophilic and thermophilic) to better understand the impact of indigenous microbial community under different start-up scenarios. With a view on solid-phase digestion, the effect of solids concentration (5–10%) was also investigated. Lastly, the solid digestate and leachate residue from a previous digestion batch were recycled to inoculate a subsequent batch, in order to test this as another means for on-going inoculation.

2. Material and methods

2.1. Raw material

Spent bedding was collected and prepared for further analysis according to Test Methods for the Examination of Composting and Compost (U.S. Composting Council, 2002) (see Sections 2.5 and 2.6 for analytical methods). Fresh samples (0–2 days old at the time of sampling) were collected from stockpiles at two piggeries (site A and B) in Queensland (Australia). The spent bedding from site A was from sheds housing smaller pigs only (called weaners, 10–24 kg), whereas spent bedding from site B was from sheds housing weaners and larger pigs (called growers, 24–36 kg). Different extents of pig exposure were visibly apparent, because the bedding samples from site A contained less faeces and urine (less soiled) than bedding samples from site B. At both sites the pigs were all reared according to a batch "all in, all out" mode. The batch time of the weaner-to-grower was 4 weeks at site A and 6 weeks at site B. At site B, the time for grower pigs to grow to slaughter weight was 3 weeks. Usually, a substantial amount of bedding straw is added at the start of the pig growth batch to cover the floor and then intermittently through the pig batch life, depending on humidity, straw durability and soilage extent. All bedding was removed at the end of each batch, usually to be stockpiled for passive composting. The average bedding use at sites A and B were about 0.30 and 0.12 kg pig^{-1} day^{-1}, respectively. Bedding at site A consisted of mixed barley straw and wheat straw (50% w/w each), and was collected for testing during winter when in-shed temperature was an estimated 20 ± 5 °C. Bedding at site B consisted of wheat straw only, and was collected during summer when in-shed temperature was an estimated 27 ± 8 °C.

2.2. Biochemical methane potential tests

Ultimate methane potential (B_0) and anaerobic biodegradability were quantified by biochemical methane potential (BMP) tests in 310 mL media bottles based on the method of Angelidaki et al. (2009). External inoculum used in the BMPs was digestate from a completely mixed mesophilic digester in South East Queensland, treating primary and secondary municipal sludge. This inoculum was added at an inoculum-to-substrate ratio (ISR) of 2 on a volatile solids (VS) basis. Bottles were flushed with high-purity nitrogen gas for 1 min at 4 L min^{-1} and were then promptly sealed with a rubber septa retained by an open top screw cap. Tests were performed in triplicate and background methane production from substrate-free blanks were subtracted. Tests were mixed by inverting once after every sampling event. Biogas volume was measured using the method described by Jensen et al. (2011) and biogas composition was determined by gas chromatography (GC) as described in Section 2.5.

2.3. Batch tests without inoculation

Batch experiments were conducted at a similar scale and using a similar method to that described above for the BMPs, except that no external inoculum was added. This was to assess the impact of indigenous microbial activity in the spent bedding samples on AD performance. The test conditions, as outlined in Table 1, included combinations of solids concentrations of 5, 10 and 20% total solid (TS), achieved by diluting the spent bedding with milli-Q® water, and test temperatures of 37 or 55 °C. Tests were performed in sextuplicate at each condition to compensate for spent bedding heterogeneity. During the course of a test set, soluble fractions were analysed by withdrawing 0.5 mL leachate samples intermittently via an 18 Gauge syringe needle. Soluble fractions were assessed from only three of the six test bottles at each test

Table 1
Test conditions of the batch experiments without external inoculum.

Spent bedding sample origin	Temperature (T, °C)	Total solids concentration (TS, %)	Test analysis performed
Site A	35	5	Gas composition and soluble content
		10	
		20	Gas composition
	55	5	Gas composition and soluble content
		10	
		20	Gas composition
Site B	35	5	Gas composition and soluble content
		10	
		20	Gas composition
	55	5	Gas composition and soluble content
		10	
		20	Gas composition

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